**Supplementary Table 1. Animal models of ACLF**

| **Hepatotoxicant** | **Animal**  **type** | **Modeling method** | | **Duration**  **required** | **Research purpose** | **Fundamental outcomes** | **Ref.** |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Chronic phase** | **Acute phase** |
| Carbon tetrachloride  (CCl4) | New Zealand  rabbit | Intraperitoneal injection of CCl4 | Intravenous injection of D-gal | 10 weeks for chronic state followed by acute phase. ACLF state last for 28 days | Evaluation of the therapeutic potential of xenogeneic porcine adipose-derived stem cells (ADSCs) in treating ACLF, and investigating the effects of porcine ADSCs transplantation on ACLF | Generating the first rabbit ACLF model transplanted with ADSCs into the livers. ADSCs transplantation prolonged the survival of ACLF animals by improving their liver function, although the underlying mechanisms are not clear | 1 |
| New Zealand  rabbit | Intraperitoneal injection of CCl4 | Intraperitoneal injection of either CCl4 or D-gal | 10 weeks for chronic state followed by acute phase. ACLF last for 12 h | To establish a large animal model of ACLF in New Zealand white rabbits for conducting further experimental targets | Among different dosages of reagents used to trigger the ACLF, this research describes the D-gal 0.7 g/kg as the ideal dose to generate ACLF over fibrosis induced by CCl4 injections | 2 |
| SD rats | Intraperitoneal injection of CCl4 | Intraperitoneal injection of D-gal combined with LPS | 10 weeks for chronic state followed by acute phase. ACLF duration is unknown | Clarifying the effects of mitofusin-2 (Mfn2) as a hepatoprotective target and explore it is mechanism of action | Mfn2 plays a protective role in the progression of ACLF through influencing multiple biological functions in ACLF via the PI3K/Akt/mTOR signaling pathway | 3,4 |
| SD rats | Intraperitoneal injection of CCl4 | Intraperitoneal injection of D-gal combined with LPS | 6 weeks for chronic state followed by acute phase. ACLF duration is 96 h | This study aimed to investigate the potential mechanisms underlying hepatoprotective effect of hypoxia-inducible factor-1α inhibitors | Genistein, a hypoxia-inducible factor-1α, could protect against ACLF by inhibiting cellular reactive oxygen species production, but the detailed mechanism is lacking | 5 |
| SD rats | Intraperitoneal injection of CCl4 | Intraperitoneal injection of D-gal combined with LPS | 8 weeks for chronic state followed by acute phase. ACLF duration is 96 h | Developing an ACLF rat model to study the dynamic changes of two immunoregulation cells, Treg and Th17 cells, during the process of ACLF | Determined by flowcytometry, the circulating trends of Treg and Th17 showed fluctuation during ACLF stage, the authors speculated that the function of Treg cell in an ACLF model was defective, but without detailed mechanisms | 6 |
| SD rats | Intraperitoneal injection of CCl4 | Intraperitoneal injection of D-gal combined with LPS | 12 weeks for chronic state followed by acute phase. ACLF duration is 24 h | To examine the protective effects of the herbal Yi-Qi-Jian-Pi formula in a rat model of ACLF | Yi-Qi-Jian-Pi formula to ACLF rats exerted protective effects against hypoxic injury which proposed through PI3K/AKT and/or RIPK1/RIPK3 pathways | 7,8 |
| SD rats | Intraperitoneal injection of CCl4 | Intraperitoneal injection of D-gal combined with LPS | 11 weeks for chronic state followed by acute phase for 4 weeks. ACLF last for 7 days | This study intended to investigate the protective effects of *San huang yin chi* decoction, derived from a well-known and canonical Chinese medicine formula, in ACLF | The study claimed a dose-dependent protective capability of the extract, possibly through promoting the APE1 / Ref-1 and regulating P53 apoptotic signaling pathway | 9 |
| C57BL6 mice | Intraperitoneal injection of CCl4 | Intraperitoneal injection of APAP followed by LPS | 10 weeks for chronic state followed by acute phase. ACLF state last for 11 days | Evaluation of the ACLF and subsequent development of portal hypertension, sepsis, and secondary organ dysfunction | This model shows the clinical and histological features of human ACLF in terms of the presence of jaundice, ascites, acute tubular necrosis, and renal dysfunction | 10 |
| C57BL/6J mice | Intraperitoneal injection of CCl4 | Intraperitoneal injection of double dose CCl4 followed by bacterial strain load or cecal ligation and puncture | 8 weeks for chronic state followed by acute phase. ACLF duration last for up to 9 days | Developing an ACLF mice model with viable bacterial infection for studying liver regeneration and exploring the therapeutic potential of interleukin-22Fc by reprogramming impaired regenerative pathways and attenuating bacterial infection | This study represents a major step forward in understanding and potentially targeting regeneration in ACLF, in which the distortion of IFNγ /STAT1 and IL-6/STAT3 pathways were responsible for liver degeneration. Interleukin-22Fc therapy promotes liver regeneration and ameliorates bacterial infection in ACLF mice | 11,12 |
| Heterologous serum: HSA or porcine serum (PS) | SD rats | Intravenous injection of HSA | Intraperitoneal injection of D-gal and LPS combination | 6 weeks for chronic state followed by acute phase. ACLF state last for 48 h | This study was established to examine the protective efficacy of recombinant adenovirus containing either single or combination of hyper-interleukin-6 and hepatocyte growth factor | The study concluded that, the protective efficacy of hyper-interleukin-6 and hepatocyte growth factor combination is more potent than that of single ones in ACLF rats, with no significant side effects observed | 13 |
| SD rats | Intraperitoneal injection of PS | Intraperitoneal injection of D-gal combined with LPS | 12 weeks for chronic state followed by acute phase. ACLF duration is 7 days | This work investigated the transcriptomic-based dataset in liver cirrhosis-based ACLF rat model developed by PS treatment | This study indicates immune-metabolism disorder in ACLF rats, in which prominent immune dysregulation at ACLF stage, whereas metabolic disruption was significantly downregulated | 14 |
| Wistar rats | Intravenous injection of HSA | Intraperitoneal injection of D-gal combined with LPS | 6 weeks for chronic state followed by acute phase. ACLF duration is 12 h | To study whether Jieduan-Niwan formula, a traditional Chinese medicine, could reduce liver apoptosis in ACLF, and the outline mechanisms | Following model establishment, the Jieduan-Niwan formula could protect against hepatic apoptosis via hindering the E2F1-mediated intrinsic apoptotic pathway | 15-18 |
| Wistar rats | Intravenous injection of HSA | Intravenous injection of D-gal and LPS combination | 6 weeks for chronic state followed by acute phase. ACLF state last for 7 days | Develop an ACLF rat model to study the role of long noncoding RNAs nuclear enriched abundant transcript 1 (NEAT1) in ACLF pathogenesis | NEAT1 over-expression reduced several cytokines expression through restriction of TRAF6 ubiquitination, however the regulation mechanism needs to be further investigated | 19 |
| Wistar rats | Intravenous injection of HSA | Intraperitoneal injection of D-gal and LPS combination | 6 weeks for chronic state followed by acute phase. ACLF duration is 12 h | Develop an ACLF rat model to study the role of soluble TNF receptor: IgG-Fc fusion protein (sTNFR:IgG-Fc) in ACLF pathogenesis | sTNFR:IgG-Fc improved survival rate, liver function and decreased inflammation and hepatocytes apoptosis, possibly via TNF/TNFRp55 blocking | 20 |
| Wistar rats | Intravenous injection of HSA | Intraperitoneal injection of D-gal and LPS combination | 6 weeks for chronic state followed by acute phase. ACLF duration is 6 h | Study the hepatoprotective potentials of the *Taraxacum officinale* ethanolic root extract on ACLF | The antioxidant activity of the plant ethanolic root extract as the potential mechanism in protection against ACLF, but no detailed mechanisms behind these protective effects | 21 |
| Wistar rats | Intravenous injection of HSA | Intraperitoneal injection of D-gal and LPS combination | 6 weeks for chronic state followed by acute phase. ACLF duration is 120 h | To examine the protective effects of Trichostatin A, which belongs to histone deacetylase inhibitors, in suppressing the inflammatory responses in ACLF | Reduced serum levels of inflammatory mediators were detected in ACLF rats received Trichostatin A, which probably through enhancing the acetylation levels of nonhistone molecules | 22 |
| Wistar rats | Intravenous injection of HSA | Intraperitoneal injection of D-gal and LPS combination | 6 weeks for chronic state followed by acute phase. ACLF state last for 7 days | The purpose of this study is to investigate the protective effects of ethyl pyruvate on ACLF rat model development | The study speculates that, ethyl pyruvate protects against ACLF through inhibiting the production of inflammatory mediators, but the detailed mechanisms are not explained | 23 |
| Wistar rats | Intravenous injection of HSA | Intraperitoneal injection of D-gal and LPS combination | 6 weeks for chronic state followed by acute phase. ACLF duration is 24 h | Whether blockade of high-mobility group box-1, an extracellular protein and inflammatory mediator, could protect against ACLF | Specific monoclonal anti- high-mobility group box-1 antibody showed protective effects against ACLF, possibly linked to interfering with TLR4 signaling pathway | 24 |
| Wistar rats | Intraperitoneal injection of PS | Intravenous injection of LPS followed by intraperitoneal injection of D-gal | 11 weeks for chronic state followed by acute phase. ACLF duration is 8 h | The purpose of this work is to determine the serum sphingolipid composition, especially the effects of dihydroceramide in ACLF | Increasing serum levels of dihydroceramide was associated with minor liver damage and preventing hepatocellular apoptosis, but no detailed mechanisms had been proposed | 25 |
| Wistar rats | Intraperitoneal injection of PS | Intravenous injection of LPS followed by intraperitoneal injection of D-gal | 11 weeks for chronic state followed by acute phase. ACLF duration is 24 h | Establish an ACLF rat model to study the effectiveness of human umbilical cord-derived mesenchymal stem cells (hUC-MSCs) in ACLF treatment | The hUC-MSC transplantation can improve liver function, liver damage and promote liver repair in ACLF, mediated most likely by inhibiting Notch signaling and reversing the imbalance of the Stat1/Stat3 pathway | 26 |
| Wistar rats | Intraperitoneal injection of PS | Intraperitoneal injection of D-gal combined with LPS | 13 weeks for chronic state followed by acute phase. ACLF duration is 12 h | To study whether Jieduan-Niwan formula, a traditional Chinese medicine, could reduce liver apoptosis in ACLF, and the outline mechanisms | Following model establishment, the Jieduan-Niwan formula could protect against hepatic apoptosis via inhibiting the JNK-induced mitochondrial apoptotic pathway | 27 |
| Wistar rats | Intraperitoneal injection of PS | Intravenous injection of LPS followed by intraperitoneal injection of D-gal | 11 weeks for chronic state followed by acute phase. ACLF duration is 48 h | Establishing a new ACLF rat model based on PS-induced liver fibrosis instead of HSA, and then exploring ACLF characteristics and mechanisms | An ACLF rat model induced by combination of PS and D-gal/LPS was established, however the underlying mechanisms of ACLF development were not fully explored | 28 |
| Surgery | SD rats | Surgically with either sham laparotomy or BDL operation | Intravenous injection of LPS | 4 weeks for chronic state followed by acute phase. ACLF duration is 3 h | Determine whether recombinant alkaline phosphatase could deactivate LPS and prevents the progression of ACLF | Recombinant alkaline phosphatase could prevent multiple organ failure development that associated with ACLF through interfering with TLR4 expression | 29 |
| SD rats | Surgically with BDL operation | Intraperitoneal injection LPS | 4 weeks for chronic state followed by acute phase. ACLF duration is 72 h | Identifying the risk of systemic inflammation in the ACLF prognosis and fatality | Elevated levels of the inflammatory cytokines, namely IL-1α and IL-1β, considered as risk factors in ACLF development form stable cirrhosis | 30 |
| SD rats or C57BL/6 mice | Surgically with BDL operation | Intraperitoneal injection LPS | 6 weeks (rats) or 14 days (mice) chronic state followed by acute phase. ACLF duration is 3 h | This study aimed to test the hypothesis that, inflammation drives the increased expression of hepatic Connexin-43 and to determine its biological role | The study outlined that, expression of hepatic Connexin-43 was related to the severity of inflammation. This increased expression is likely to be an adaptive protective response of the liver to facilitate cellular communications | 31 |

HSA-based model, animals were subcutaneously administrated with repeated injection of Freund’s adjuvant containing HSA on days 0, 14, 24, and day 34. Ten days after the last injection, the concentration of serum HSA from these immunized rats was detected to confirm the sensitized status. After that, these sensitized rats were injected with intravenous HSA to develop chronic liver disease status.

ACLF, acute-on-chronic liver failure; ADSCs, adipose-derived stem cells; AKT, protein kinase B; APAP, acetaminophen; APE1, apurinic/apyrimidinic endonuclease; BDL, bile duct ligation; C57BL6, C57 black 6; C57BL/6J, C57 black 6/J strain; CCl4, carbon tetrachloride; D-gal, galactosamine; E2F1, E2F transcription factor 1; HSA, human serum albumin; hUC-MSCs, human umbilical cord-derived mesenchymal stem cells; IL-1α, interleukin 1 alpha; IL-1β, interleukin 1 beta; IL-6, interleukin 6; IFNγ, Interferon gamma; IgG-Fc, immunoglobulin G-Fc fusion protein; JNK, c-Jun N-terminal kinase; LPS, lipopolysaccharide; Mfn2; mitofusin-2; mTOR, mammalian target of rapamycin; NEAT1, nuclear enriched abundant transcript 1; P53, tumor protein P53; PI3K, phosphoinositide 3-kinase; PS, porcine serum; Ref-1, redox effector factor-1; RIPK1, receptor-interacting serine/threonine-protein kinase 1; RIPK3, receptor-interacting serine/threonine-protein kinase 3; RNAs, ribonucleic acids; SD, Sprague-Dawley; STAT1, signal transducer and activator of transcription 1; STAT3, signal transducer and activator of transcription 3; sTNFR, soluble tumor necrosis factor receptor; sTNFR:IgG-Fc, soluble tumor necrosis factor receptor: immunoglobulin G-Fc fusion protein; Th17, interleukine-17-producing helper T cells; TLR4, toll-like receptor 4; TNF, tissue necrotic factor; TNFRp55, tumor necrosis factor receptor superfamily, member 1a; TRAF6, tissue necrotic factor (TNF) receptor associated factor 6; Treg, regulatory T cells.

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